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From fresh to dry: Moth body weight changes through preservation

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Abstract. Body weight is a key trait in insects, reflecting size, condition, and energetic status. It also influences their flight performance, reproduction, and ecological interactions. Yet most ecological and taxonomic studies rely on preserved rather than living individuals, despite the substantial mass loss caused by drying and other curatorial procedures. We quantified how preservation affects body mass in moths (Lepidoptera) using three complementary datasets representing major workflows: laboratory-reared *Spodoptera littoralis* (Boisduval, 1833), freshly collected moths from Germany, and frozen geometrid specimens from Peru. Across datasets, body weight declined markedly across developmental stages and preservation states, with only partial recovery after rehydration. In *S. littoralis*, females were consistently heavier than males, while egg presence did not influence female mass. By integrating results across workflows, we derived practical correction factors for estimating fresh body weight from dry specimens. These order-level approximations substantially improve the ecological value of preserved material by allowing body-mass data from museum collections to reflect the mass of living insects more accurately. Applying such corrections enhances the reliability of trait-based and comparative analyses that depend on preserved Lepidoptera.

Introduction

Body weight is a fundamental trait in insects, reflecting size, physiological condition, energetic reserves, and ecological performance (Chown and Gaston 2010; Xu and Wang 2020). In moths (Lepidoptera), body mass varies across developmental stages, sex, diet, and species (Ojeda-Avila et al. 2003; Molleman et al. 2011), and is further shaped by preservation methods. Drying, for example,

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removes water and other volatile compounds from tissues, causing shrinkage and increased fragility (Gilbert 2011; Sheikh et al. 2017). Desiccation can also collapse membranous structures, alter protein conformation, and change tissue density and texture, potentially distorting the actual body proportions and reducing the accuracy of morphological or mass-based measurements (Chown et al. 2011; Gilbert 2011). It additionally modifies the physical properties of the cuticle, affecting structural stability and hydration capacity, with measurable consequences for external morphology (Fraenkel and Rudall 1947; Wipfler et al. 2016). Furthermore, in Lepidoptera, drying has been shown to alter wing length and area, potentially biasing morphometric measurements (Van Hook et al. 2012). Consequently, preservation history should be considered when interpreting downstream analyses, particularly in studies where dry body weight is assumed to reflect live mass or physiological state.

Fresh weight of a winged insect represents the functional mass most linked to flight energetics, reproductive investment, and overall condition (Chown and Gaston 2010). It generally provides a more informative measure than dry weight or structural proxies, which may overlook variation in hydration and lipid content (Abdel-Maksoud et al. 2011; García-Barros 2015). Across Lepidoptera, fresh weight often outperforms linear measurements or dry mass as an indicator of body condition and dispersal capacity (Beck and Kitching 2007; García-Barros 2025), and it is essential for calculating functional traits such as wing loading (Correa-Carmona et al. 2022). However, fresh weight is rarely measured in large-scale biodiversity surveys or tropical fieldwork, and preserved specimens constitute the primary source of body-mass data for ecological and museum-based research (Gilbert 2011; Wardhaugh 2013).

Insects lose water rapidly after death, with smaller species desiccating especially quickly owing to their high surface-to-volume ratio (Noble-Nesbitt 1969; Sample et al. 1993; Molleman et al. 2011). Additional mass loss may occur during freezing, thawing, mounting, and long-term storage (Sanford et al. 2011; Bogusz et al. 2024). Although rehydration is widely used to soften dry specimens for preparation (Walker and Crosby 1988; Krogmann and Holstein 2010), rehydrated specimens do not recover their original live (fresh) weight, and most regained moisture is lost again during subsequent drying (Thorat and Nath 2018; Neisskenwirth 2020). As a result, body-weight values derived from preserved adult specimens primarily reflect dry weight influenced by specimen handling procedures rather than fresh(live) body weight, introducing uncertainty when such measurements are interpreted as proxies for physiological condition or biomass (Moretti et al. 2017). This emphasises the importance of documenting specimen handling history (e.g. drying, rehydration, storage conditions) and developmental stage (e.g. freshly emerged vs. worn adults) in trait databases (Keller et al. 2023).

In this study, we quantify body weight changes across developmental and preservation stages using three complementary datasets that reflect common curatorial and ecological practices. The first consists of laboratory-reared *Spodoptera littoralis* (Boisduval, 1833) measured at pupal, live adult, and dead dry adult stages, including sex-specific variation and the influence of egg presence. The second comprises freshly collected moths from Germany weighed before and after air-drying, representing natural field-to-laboratory workflow. The third includes geometrid moths from Peru that were frozen, rehydrated, mounted, and allowed to dry, which corresponds to a typical workflow from fieldwork to the prepared insect in a museum's insect drawer. Because no single dataset spans all stages, combining them allows us to reconstruct a full sequence from live fresh adults to dry mounted specimens.

Although the taxa and contexts differ, the primary drivers of mass change (e.g., water loss, lipid depletion, and structural modification) are shared broadly across Lepidoptera (Noble-Nesbitt 1969;

Chown and Gaston 2010). Our aim is therefore not to generalise physiological mechanisms across species, but to provide practical, order-level correction factors that allow researchers to better estimate fresh body weight from preserved moth specimens, thereby enhancing the ecological and functional value of museum collections.

Materials and methods

We measured body weight using a Sartorius BP121S precision balance (readability 0.001 g, or 0.1 mg). We analysed three datasets that differed in sampling design and specimen treatment but shared a common analytical framework.

Life cycle dataset (laboratory)

We reared 81 individuals of *Spodoptera littoralis* (Lepidoptera: Noctuidae) on an artificial diet provided by the Max Planck Institute for Chemical Ecology (Jena, Germany). We maintained all larvae under identical laboratory conditions in September 2024 at 20–24°C and 40–50% relative humidity.

We weighed pupae at least 24 hours after pupation. Adult emergence occurred predominantly at night; we weighed adults the following morning, after complete wing expansion and meconium excretion, to obtain live adult weight from newly emerged, unfed individuals. We then kept adults without food until natural death. After death, we air-dried individuals for 10 days and weighed them again to obtain dry adult weight. Live and dry adult weights represent distinct physiological states of the adult stage, and do not constitute repeated measurements of the same condition. In none of our experiments we did apply oven-drying because it is not common practice in insect curation. Our aim was to quantify body mass changes under realistic curatorial and laboratory conditions, rather than to define an absolute dry mass standard. We recorded sex for all *Spodoptera* individuals. In females, we assessed egg presence by direct observation of eggs laid prior to death and classified females that did not lay eggs as lacking visible egg deposition.

Field-collected dataset (Germany)

We collected 71 moth individuals belonging to 13 families at the Naturschutzstation Dr. Helmut Steuer, Unteres Schwarzatal, Thuringia, Germany in July 2024 (50.668°N, 11.222°E (DD)). Six specimens could not be reliably identified to family level because of poor condition and are therefore reported as N/A.

We collected all specimens alive. After death, we stored specimens in a sealed container maintained at near-saturated humidity (~100%) to minimise post-mortem desiccation. We weighed specimens the following morning to obtain fresh dead weight. We then air-dry specimens for 10 days under laboratory conditions (20–24 °C, 40–50% relative humidity) and weighed them again to obtain dry weight.

Rehydration dataset (Peru)

We analysed 30 geometrid moth specimens collected in Peru in 2023 as part of the ANDIV project (Holzmann et al. 2025). Specimens had died shortly after capture, been allowed to dry, and were subsequently stored frozen at -18°C in glassine bags inside plastic containers. Specimens were transported to Germany under dry conditions and stored at the Phyletisches Museum (Jena, Germany), where they remained frozen at -18°C for several months prior to analysis.

We weighed specimens in September 2024 under laboratory conditions (20–24 °C, 40–50% relative humidity). We measured body weight at three preparation stages: (1) frozen weight, measured immediately after removal from frozen storage; (2) rehydrated weight, measured after rehydration; and (3) prepare dry weight, measured ten days after mounting and air drying.

We rehydrated specimens in a sealed plastic container at approximately 100% relative humidity for 24 hours following standard protocols (Walker and Crosby 1988; Krogmann and Holstein 2010), allowing specimens to become sufficiently flexible for handling and mounting. Rehydrated weight was recorded at this stage, as specimens are commonly handled and weighed during rehydration in museum workflows. Prior to mounting, we recorded the weight of the mounting pin and subtracted it from the final prepared (dry) weight.

Statistical analysis

We conducted all statistical analyses in R v4.4.0 (R Core Team 2023). To avoid pseudoreplication arising from repeated measurements of the same individuals, we analysed body weight using linear mixed-effects models, with individual identity included as a random intercept in all datasets.

We analysed body weight on the log-transformed scale, which improved normality and homoscedasticity of model residuals. For each dataset, we included the relevant weight state as a fixed effect:

(1) life stage and adult condition (pupa, live adult, dry adult) in the laboratory dataset;
(2) preservation condition (fresh dead adult vs dry dead adult) in the German field dataset; and
(3) specimen preparation stage (frozen, rehydrated, prepared dry) in the Peruvian dataset.

Where applicable, we included additional fixed effects (sex, egg presence, or family).

We fitted models using restricted maximum likelihood (REML) and assessed the significance of fixed effects using Type III ANOVA with Satterthwaite or Kenward–Roger approximations for degrees of freedom. To quantify differences among levels of fixed effects, we estimated model-based marginal means and performed Tukey-adjusted pairwise comparisons. To estimate fresh or rehydrated body weight from dry specimens, we calculated correction factors using two complementary approaches across datasets. We first quantified the mean ratio between weights measured under different conditions and estimated uncertainty using non-parametric bootstrap confidence intervals. We then fitted regression-based calibration models relating log-transformed weights measured under different conditions to assess size dependence and quantify prediction uncertainty.

We produced all figures using ggplot2 (Wickham 2016), with individual-level data overlaid on boxplots to visualise variation across stages, conditions, and treatments. Analyses used the R packages lme4/lmerTest (Kuznetsova et al. 2017), emmeans (Lenth 2023), dplyr (Wickham et al. 2019), patchwork (Pedersen 2020), and reshape2 (Wickham 2007).

Results

Developmental weight changes in *Spodoptera littoralis*

The dataset included 81 individuals, comprising 35 females (43%) and 46 males (57%). Body weight differed markedly among developmental and physiological stages (Fig. 1).

Pupal weight ranged from 0.190 to 0.419 g (mean=0.268 g, SD = 0.044). Live adult weight ranged from 0.071 to 0.247 g (mean = 0.137 g, SD = 0.033), while dry adult weight ranged from 0.022 to

0.109 g (mean = 0.041 g, SD = 0.015). Thus, individuals lost substantial weight during metamorphosis and experienced an additional pronounced reduction in body weight after death and drying (Table 1). Females consistently weighed more than males across all stages. Female pupae averaged 0.281 g, compared to 0.259 g in males. Live adult females averaged 0.154 g, whereas males averaged 0.124 g. After drying, females retained higher weight (0.047 g) than males (0.036 g). These descriptive patterns indicate strong effects of developmental stage and sex on body weight (Fig. 1; Table 1).

Mixed-effects models confirmed these patterns statistically (see Suppl. material 1: table S1). Developmental stage had a strong effect on body weight ($p < 0.001$), and sex also significantly affected weight ($p = 0.001$), with a significant Stage × Sex interaction ($p = 0.001$; Table 2). In contrast, egg presence did not significantly influence body weight and did not interact with developmental stage ($p > 0.5$).

Estimated marginal means showed a consistent ranking of stages in both sexes, with pupae being heaviest, followed by live adults and then dry adults. All pairwise comparisons among stages within each sex were statistically significant (Tukey-adjusted $p < 0.001$; Suppl. material 1: table S2).

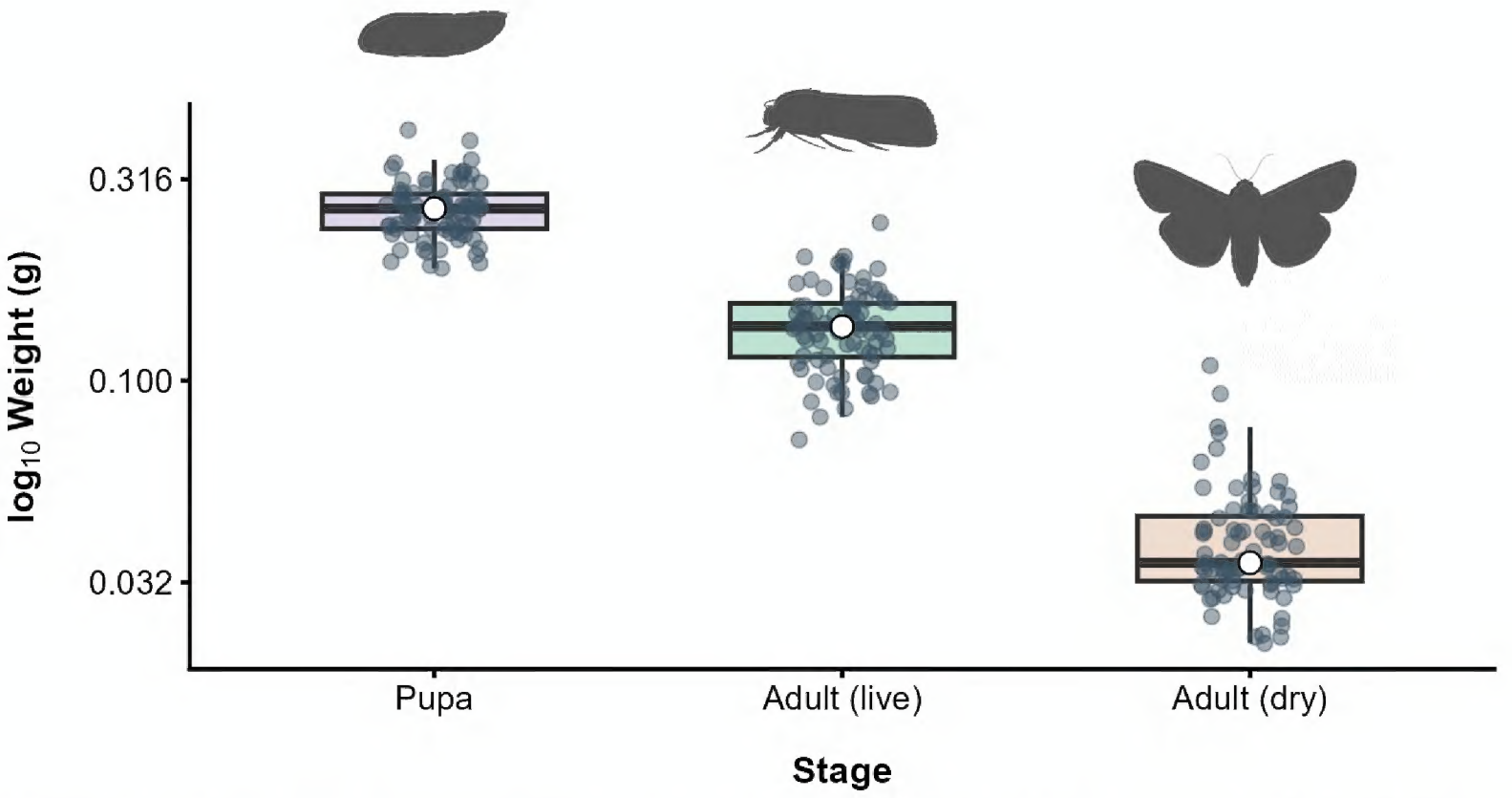

Figure 1. Body weight (log_{10} g) of *Spodoptera littoralis* pupae, live adults, and dry adults. Points show individual measurements; boxplots indicate medians and interquartile ranges, and white circles show means.

Table 1. Sample composition and mean body weight (g = grams; ± SD = standard deviation) by sex and life stage in *Spodoptera littoralis*.

	Group	n (%)	Pupal weight (g)	Live adult weight (g)	Dry adult weight (g)
Female (F)	All	35 (43%)	0.2808 ± 0.0416	0.1540 ± 0.0332	0.0471 ± 0.0172
	With eggs	17	–	0.1556 ± 0.0391	–
	Without eggs	18	–	0.1525 ± 0.0276	–
Male (M)	–	46 (57%)	0.2585 ± 0.0443	0.1241 ± 0.0266	0.0357 ± 0.0104
Total	–	81 (100%)	0.2681 ± 0.0443	0.1372 ± 0.0330	0.0407 ± 0.0148

Table 2. Type III ANOVA for the linear mixed-effects model, response: log (Weight). Satterthwaite approximation used for denominator degrees of freedom.

Effect	Sum Sq	Mean Sq	NumDF	DenDF	F value	p
Stage	94.456613397	47.228306699	2	153.96	2,069.34	<0.001
Sex	0.258768472	0.258768472	1	77.58	11.34	0.001
Eggs	0.005406531	0.005406531	1	77.41	0.24	0.628
Stage:Sex	0.321420183	0.160710091	2	153.97	7.04	0.001
Stage:Eggs	0.026788390	0.013394195	2	153.80	0.59	0.557

Regression-based calibration further showed a strong positive relationship between preserved and live adult weight ($p < 0.001$). The slope of this relationship was significantly less than one, indicating size-dependent mass loss, with smaller individuals losing proportionally more mass during preservation. Sex and egg presence did not significantly affect this relationship (Table 3).

Drying effects across taxonomic groups

Fresh dead body weight ranged from 0.0078 to 0.421 g (mean = 0.085 g, SD = 0.092), while dry body weight ranged from 0.0026 to 0.183 g (mean = 0.033 g, SD = 0.038). Drying therefore resulted in a substantial reduction in body mass across all specimens (Fig. 2).

Table 3. Regression-based calibration model predicting live body weight from preserved weight. Linear model fitted on log-transformed weights. Alive = live adult weight; Dead = preserved (dry) adult weight. Neither sex nor egg presence significantly affected the relationship.

Predictor	Estimate	SE	t	p
(Intercept)	-0.457	0.227	-2.02	0.047
Log (Dead)	0.464	0.072	6.47	<0.001
Sex M	-0.086	0.052	-1.66	0.100
Eggs Yes	0.005	0.059	0.08	0.934

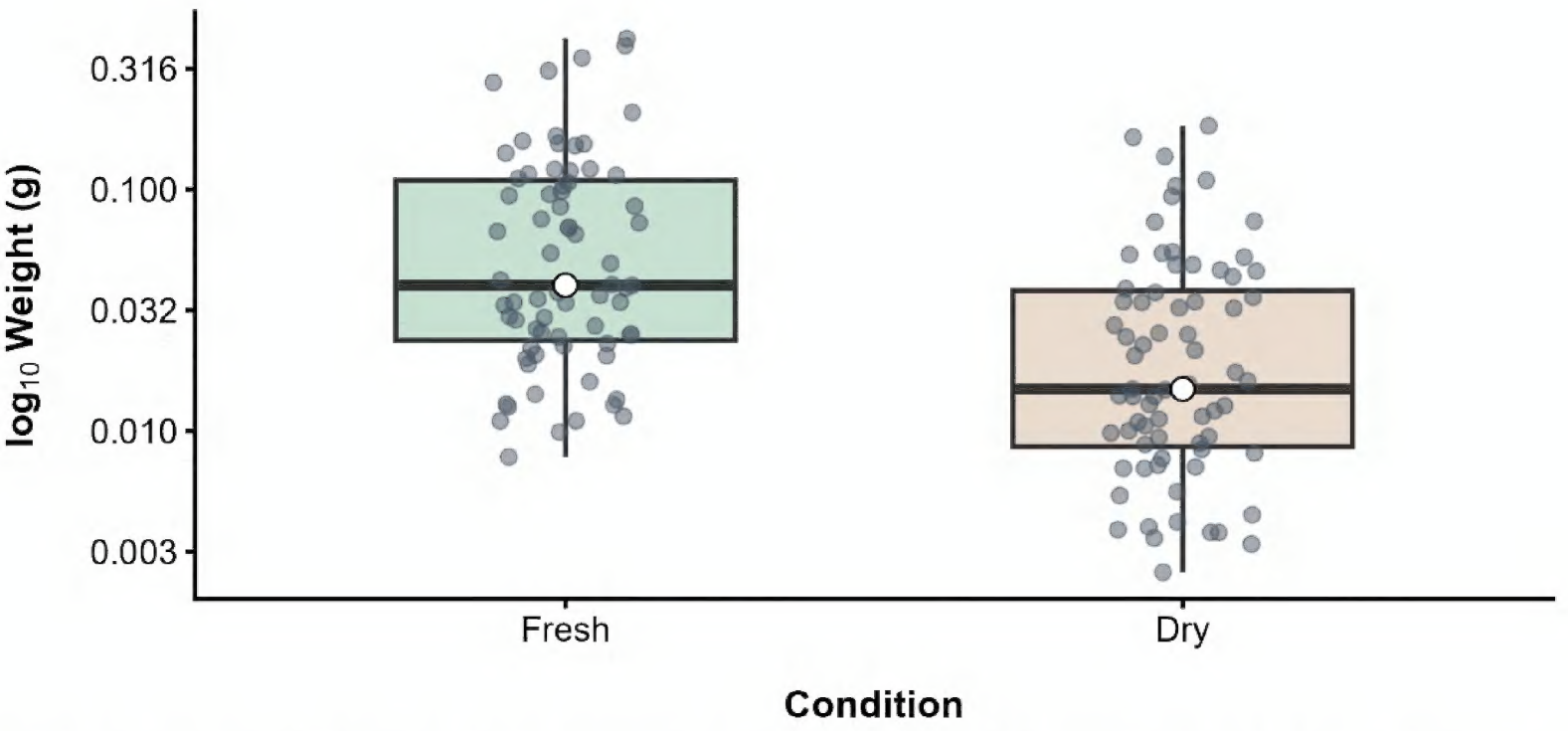

Figure 2. Body weight ($\log_{10}$ g) of field-collected moths measured as fresh and dry weight. Points show individual specimens; boxplots indicate medians and interquartile ranges, and white circles show means.

Mixed-effects models accounting for paired measurements within individuals confirmed a strong effect of measurement condition on body weight. Dry specimens weighed significantly less than fresh specimens (estimate = −1.01 on the log scale, $p < 0.001$; Table 4), corresponding to an average reduction of approximately two-thirds of fresh mass.

Estimated marginal means showed a clear separation between fresh and dry conditions across specimens (Table 5), with dry body weight consistently lower than fresh body weight.

Descriptive summaries showed substantial variation in absolute body weight among families in both fresh and dry conditions (Suppl. material 1: table S3). These differences reflect variation in overall body size across taxonomic groups.

When family was included in the analysis, several family-level estimates differed from the reference level (Suppl. material 1: table S4). However, the reduction in body weight from fresh to dry condition was consistent across families, and no family-specific drying effects were identified. Family-level results are therefore reported descriptively.

Effects of freezing, rehydration, and preparation on body weight

Body weight differed markedly among preparation stages (Fig. 3). Rehydrated specimens showed higher body weight than both frozen and prepared dry specimens, whereas prepared dry specimens consistently had the lowest weights.

Table 4. Mixed-effects model (Condition effect).

effect	term	estimate	std.error	statistic	df	p.value
fixed	ConditionDry_Weight	-1.006	0.019	-53.52	70	<0.001

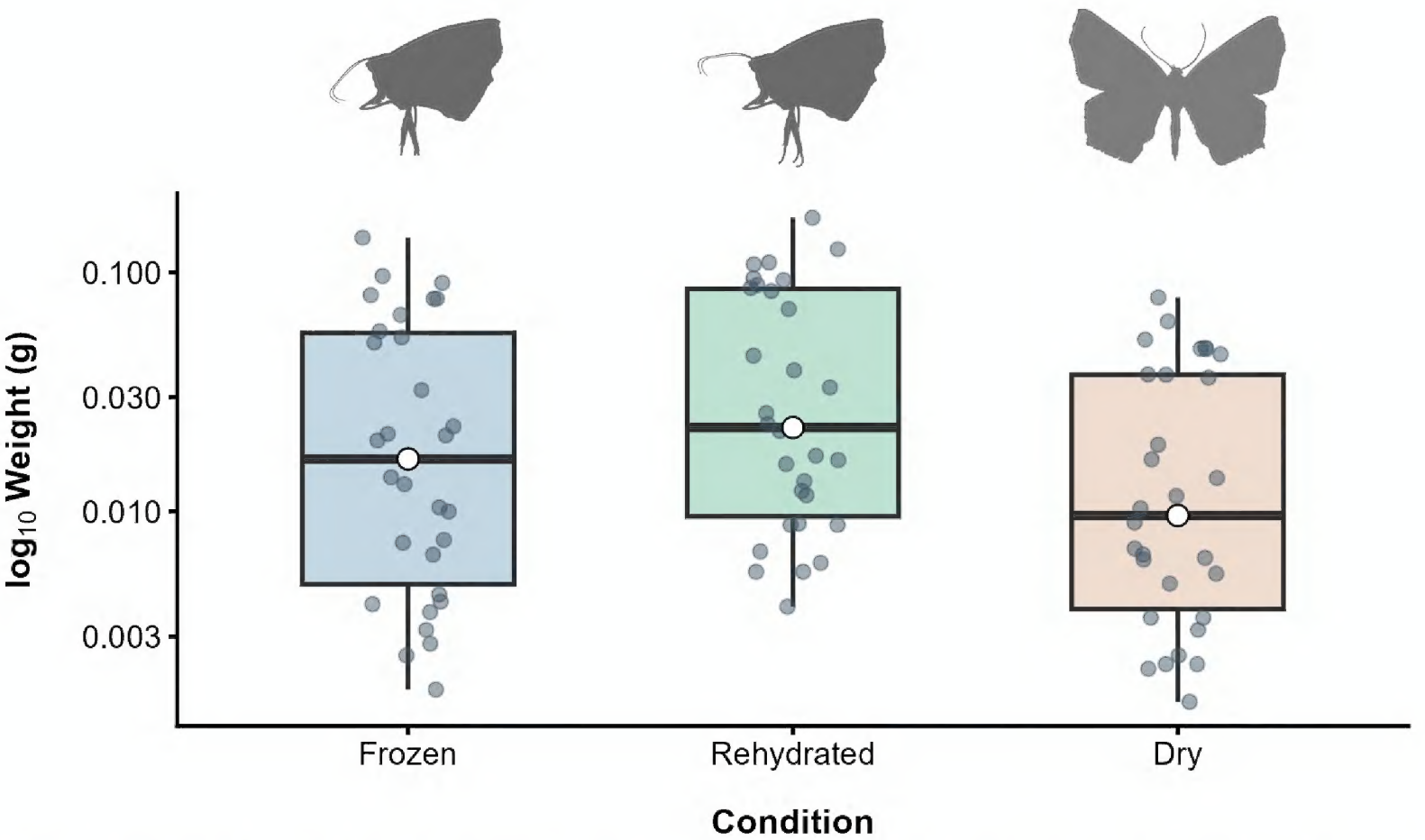

Figure 3. Effects of freezing, rehydration, and preparation on body weight of Peruvian geometrid moths. Points represent individual specimens; boxplots show medians and interquartile ranges, with whiskers indicating 1.5× the interquartile range. White circles indicate mean values.

Table 5. Estimated marginal means (Fresh vs Dry).

Condition	emmean	SE	df	lower.CL	upper.CL
Wet_Weight	-2.980	0.125	57.64489	-3.231	-2.729
Dry_Weight	-3.986	0.125	57.64489	-4.237	-3.735

Table 6. Mixed-effects model testing the effect of preparation stage on body weight.

Effect	Estimate	SE	df	t	p
(Intercept)	-4.106	0.222	29.42778	-18.49	<0.001
Rehydrated_weight	0.449	0.033	58.00000	13.65	<0.001
Dry_weight	-0.386	0.033	58.00000	-11.73	<0.001

Mixed-effects models confirmed a strong effect of preparation stage on body weight (Table 6). Estimated marginal means revealed a consistent ranking across specimens, with rehydrated weight highest, followed by frozen weight and prepared dry weight lowest (Suppl. material 1: table S5). All pairwise differences among preparation stages were statistically significant.

Correction factors for estimating live, fresh, or rehydrated body weight from dry specimens

Across all three datasets, dry specimens consistently underestimated body weight relative to live, fresh, or rehydrated conditions. Correction factors derived from paired measurements showed clear and reproducible patterns, with relatively low uncertainty.

In the laboratory dataset, live adult body weight was on average 3.53 times higher than dry adult mass (SD = 0.78), based on paired measurements within individuals. Bootstrap resampling produced a narrow 95% confidence interval of 3.36–3.70 (Table 7). Sex and egg presence had negligible effects on this relationship. A simplified correction equation can therefore be used to estimate live body weight from dry specimens. When uncertainty needs to be quantified, regression-based predictions with confidence intervals are recommended. The most parsimonious correction equation was: $Live = exp(-0.457) \times dry\text{^}0.464$.

In the German field dataset, fresh dead body weight was on average 2.77 times higher than dry body mass (SD = 0.44; n = 71). Bootstrap resampling yielded a 95% confidence interval of 2.67–2.87 (Table 7), indicating consistent mass loss during drying. Regression analysis of log-transformed weights revealed a nearly proportional relationship between fresh and dry mass (slope = 0.95, intercept = 0.79, $R^2 = 0.98$, $p < 0.001$). The slope was slightly below one ($\beta = 0.92$), indicating weak size dependence. No significant differences among families were detected ($F_{10.47} = 1.23$, $p = 0.30$). A single correction equation therefore describes the relationship across families: $Fresh = exp(0.788) \times dry\text{^}0.946$.

Table 7. Summary of correction factors across datasets.

Dataset	Comparison	n	Mean ratio	SD	95% CI
Laboratory (*Spodoptera*)	Live/dry	81	3.53	0.78	3.36–3.70
Field (Germany)	Fresh/dry	71	2.77	0.44	2.67–2.87
Rehydration (Peru)	Rehydrated/ dry	30	2.32	0.24	2.23–2.40

In the Peruvian geometrid moth dataset, rehydrated body weight was on average 2.32 times higher than prepared dry mass (SD = 0.24; n = 30). Bootstrap resampling produced a 95% confidence interval of 2.23–2.40 (Table 7), indicating low uncertainty. Regression analysis showed a strong relationship between prepared dry and rehydrated weight ($R^2 = 0.996$, $p < 0.001$). The slope ($\beta = 0.93$) was close to one, again indicating weak size dependence. The corresponding correction equation was: $Rehydrated = exp(0.537) \times dry\text{^}0.934$.

Discussion

Dry preservation substantially reduces measured body weight in moths, such that dry weight represents only a fraction of live or fresh mass. Because this study is methodological in scope, we focus on quantifying the effects of preservation on moth body weight and on providing correction factors that allow live, fresh, or rehydrated weight to be estimated from preserved specimens.

Across the three complementary datasets, body mass followed a consistent pattern: marked reductions during metamorphosis and desiccation, and only partial recovery after rehydration. These results demonstrate that preserved measurements alone can substantially underestimate the mass of living insects. Although biological variation was evident, particularly sexual dimorphism, weight loss trajectories were highly consistent across taxa and preservation workflows, supporting the use of standardised correction approaches to reconstruct live or fresh weight from preserved material.

Developmental changes from pupa to adult

In *Spodoptera littoralis*, body mass declined by approximately half between the pupal and adult stages, reflecting water loss, meconium excretion, and shedding of the pupal case during metamorphosis (Jungreis et al. 1982; Molleman et al. 2011). Subsequent desiccation reduced adult mass by an additional ~70% from live to dry adult, resulting in an overall loss of ~85% from pupa to dry adult. These findings underscore the extent to which both developmental processes and specimen preparation influence body weight in Lepidoptera (Chown et al. 2011; Gilbert 2011; Bogusz et al. 2024). The pupal–adult weight relationship is treated as species-specific and exploratory and is restricted to the laboratory-reared *S. littoralis* dataset; it is not used for correction factors or extrapolation across species.

Sexual dimorphism in body mass, with females consistently heavier than males, aligns with patterns documented across multiple Lepidoptera lineages (Allen et al. 2011; Stillwell et al. 2014; De Camargo et al. 2015). Egg presence did not result in higher female weight, suggesting that *S. littoralis* allocates most reproductive resources gradually after emergence. This pattern is consistent with an income-breeding strategy, in which adult feeding and ongoing resource acquisition contribute substantially to egg production (Tammaru and Haukioja 1996; Javoiš et al. 2011; Teder 2014; Davis et al. 2016), in contrast to capital-breeding strategy of groups such as Saturniidae with non-feeding adults (Correa-Carmona et al. 2022).

Weight loss due to drying

Freshly collected moths from Germany lost, on average, approximately 63% of their fresh mass after air-drying. This magnitude of mass loss is consistent with previous reports of water loss following insect death (Noble-Nesbitt 1969; Benk et al. 2020). The resulting ratio, with dry weight representing about 37% of fresh weight, closely matches estimates reported for Lepidoptera by

García-Barros (2025). Our regression analyses further showed that dry weight strongly predicted fresh weight, with only weak size dependence and no detectable family-level effects, indicating that a single correction equation adequately describes the drying effect across taxa included in this dataset.

These results highlight why uncorrected dry weights underestimate energetically and functionally relevant traits such as energy reserves, wing loading, and flight capacity (Rogers et al. 1976; Sample et al. 1993; García-Barros 2015).

Rehydration and partial mass recovery

In the Peruvian dataset, rehydration increased specimen weight by approximately 61% relative to prepared dry mass, yet rehydrated individuals remained substantially lighter than their estimated fresh condition. Rehydrated moths therefore represented only a partial recovery of pre-drying mass (Sanford et al. 2011; Thorat and Nath 2018). Regression analyses showed that prepared dry weight was an excellent predictor of rehydrated weight, with near-proportional scaling and minimal size dependence.

Incomplete recovery probably reflects limited water infiltration into internal tissues and irreversible structural changes caused by desiccation. During drying, intracellular water loss can collapse muscle fibres and damage lipid membranes, reducing porosity and altering capillary action (Abdel-Maksoud et al. 2011; Chown et al. 2011; Sheikh et al. 2017). Consequently, rehydration primarily affects outer cuticular layers and hemolymph spaces rather than deeply embedded tissues (Oliver et al. 2020; O'Donnell 2022). Frozen specimens may retain residual moisture, contributing to greater structural stability and more moderate mass loss compared to fully air-dry individuals (Neisskenwirth 2020).

We do not interpret rehydrated body weight as an estimate of live or fresh body mass. Instead, it represents a workflow-specific reference stage, as rehydration is routinely applied prior to mounting museum specimens. At this stage, specimens are less fragile than when fully dry, making weighing more practical while acknowledging its biological limitations.

Workflow integration and correction factors

Each dataset represents a common preservation workflow in Lepidoptera research, including laboratory rearing, fresh field collection, and museum specimen preparation (Walker and Crosby 1988; Krogmann and Holstein 2010; Neisskenwirth 2020). Analysed together, these datasets allow reconstruction of a realistic sequence of body-mass change from fresh or live adults through drying and subsequent rehydration (Sanford et al. 2011; Bogusz et al. 2024).

Across workflows, preservation consistently resulted in substantial mass loss (Fig. 4). Dry specimens retained approximately 30–40% of fresh or live body mass, whereas rehydrated specimens recovered only part of this loss, reaching approximately 60–65% of fresh mass.

Regression-based calibration showed that mass change during preservation followed predictable scaling relationships. In the laboratory dataset, regression slopes below one indicated size-dependent mass loss, with smaller individuals losing proportionally more mass than larger individuals. In contrast, slopes in the German and Peruvian datasets were close to one, indicating largely proportional mass change across specimen sizes. Despite this variation in size dependence, all workflows exhibited strong and systematic relationships between dry and fresh or live body mass. This suggests that preservation-induced water loss and partial water uptake during rehydration follow systematic and quantifiable patterns across taxa and preparation methods, consistent with previous studies on mass change in preserved insects (Sanford et al. 2011; Bogusz et al. 2024).

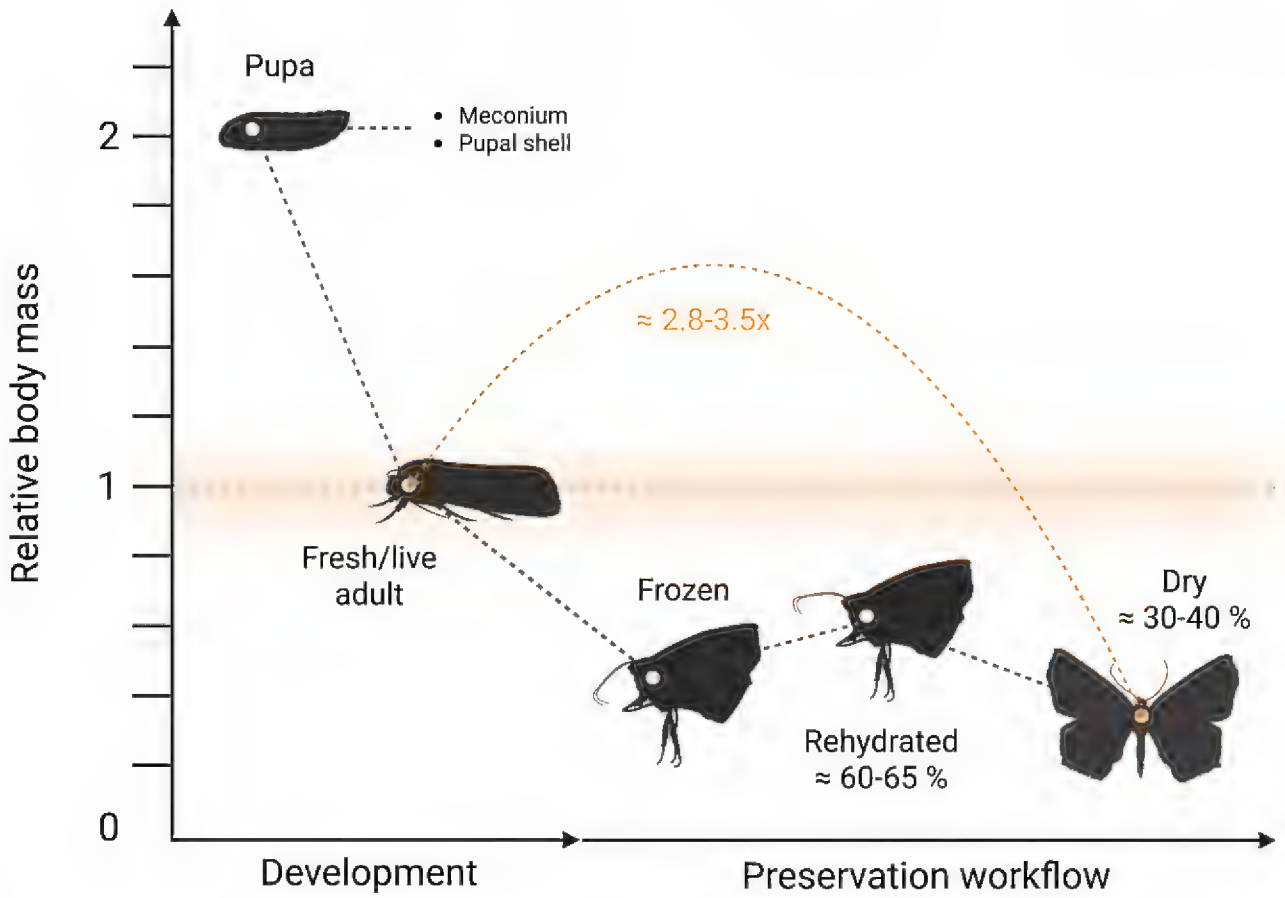

Figure 4. Conceptual diagram illustrating changes in body mass from fresh (live) adult Lepidoptera to dry preserved specimens and after rehydration. Dry specimens retain approximately 30–40% of fresh (live) body mass, whereas rehydrated specimens recover approximately 60–65%. Arrows indicate mean multiplicative correction factors derived from paired measurements (fresh/live to dry ≈ 2.8–3.5×).

Fig. 4 integrates these results into a conceptual framework linking developmental transitions and preservation workflows. The diagram illustrates the average multiplicative correction factors derived from paired measurements, showing that fresh or live adult mass can be estimated from dry specimens using correction factors of approximately 2.8–3.5×, while rehydrated mass can be estimated from prepared dry specimens using a factor of approximately 2.3×. Together, these results demonstrate that preservation-induced mass loss and partial mass recovery during rehydration follow consistent and quantifiable patterns across Lepidoptera, supporting the use of standardized correction factors to approximate fresh or live body weight from preserved material.

Ecological implications

Accurate estimates of body weight are central to insect ecology, as mass influences a broad range of physiological, behavioural, and life-history traits (Peters 1986; Kalinkat et al. 2015). Fresh weight best reflects the functional mass available for flight, metabolism, and reproduction, and is therefore essential for estimating traits such as wing loading, metabolic rate, and dispersal capacity (Beck and Kitching 2007; Boggs 2009). Body weight also integrates environmental and nutritional influences, including larval diet quality, adult feeding behaviour, and responses to temperature and humidity (Chown and Gaston 2010; Molleman et al. 2011; Padda and Stahlschmidt 2022).

Because fresh specimens are rarely available for large-scale surveys or museum-based studies, correction factors are essential to avoid systematic underestimation of ecologically relevant traits (Rogers et al. 1976; Sample et al. 1993; García-Barros 2025). Our results show that

preservation can reduce body mass by more than half, highlighting the magnitude of bias introduced when dry weight is used uncorrected. Applying correction factors, reporting specimen condition, and documenting preservation workflow will substantially improve the comparability and reliability of trait-based analyses (Chown and Gaston 2010; Molleman et al. 2011; Gallagher et al. 2020; Keller et al. 2023).

Limitations

A fully continuous dataset spanning all developmental and preservation stages is logistically unfeasible for most Lepidoptera. Our multi-dataset approach therefore represents a practical compromise that captures the principal sources of body weight variation using workflows that reflect common field and curatorial practices. Specimens were air-dry under laboratory conditions rather than oven-dry, which may allow some residual moisture and limits direct comparability with studies using highly standardized drying protocols at high temperatures.

The correction factors presented here should be considered as order-level approximations rather than species-specific physiological models. They are intended to improve the interpretation of preserved specimens and do not replace direct measurements of fresh weight.

Conclusions

Body weight in Lepidoptera is strongly shaped by both metamorphosis and specimen preservation, with preserved specimens retaining only a fraction of their original fresh mass. Although the magnitude of mass loss varies among developmental stages and preservation workflows, these changes are consistent and predictable.

By integrating laboratory, field, and museum datasets that represent common curatorial workflows, we provide practical correction factors that enable fresh or live body weight to be estimated from dry specimens. Accounting for preservation-related mass change will improve the comparability of studies across collections and time and enhance the use of Lepidoptera body weight as a functional trait in ecological and evolutionary research.

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Supplementary material 1

Supplementary tables S1–S5

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Data type: docx

Explanatory note: **table S1:** Linear mixed-effects model fixed effects. Estimates are on the log scale; **table S2:** Tukey-adjusted pairwise comparisons of developmental stages within egg presence. P-values are adjusted using the Tukey method. Estimates are on the log scale; **table S3:** Descriptive summary of fresh and dry body weight by family. Values show mean body weight (g) with standard deviation in parentheses; **table S4:** Mixed-effects model coefficients for family; **table S5:** Estimated marginal means of body weight by preparation stage.

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